

Supplement: 1: Anti-SRP and anti-synAbs were detected using a combination of protein and RNA IP. Briefly, a 20 μ l serum sample was bound overnight at 4°C to 2 mg Protein A Sepharose CL-4B beads (Amersham Biosciences, Piscataway, NJ) and washed 3 times with IP buffer (10mM Tris/HCl pH 8.0, 500 mM NaCl, 0.1% Igepal CA630). For protein IP, the IgG bound Protein-A Sepharose was then resuspended in 500 μ l of IP buffered 35 S methionine labeled extract from approximately 1×10^6 rapidly dividing K562 cells and incubated 2 hours at 4°C. The beads were washed 3 times with IP buffer, suspended in 2x Laemmli sample buffer, loaded on a standard size 8% gel, and electrophoresed at 200 V. The gel was enhanced with 0.5M sodium salicylate, dried, and autoradiographed for 3-6 days. Apparent molecular weights were determined by comparison with known 14 C labeled standards run concurrently.

For RNA IP, the IgG bound Protein-A Sepharose was resuspended in 300 μ l NET-2 buffer (50mM Tris/HCl, pH 7.4, 150 mM NaCl, 0.05% Igepal CA630), and incubated with 200 μ l K562 whole cell extract/NET-2 buffer for 2 hours at 4°C. After 3 washes with NET-2 buffer, the resultant complexes were resuspended in 350 μ l extraction buffer (NET-2, 0.25 M sodium acetate, 0.83% SDS, 1 μ l glycogen) and extracted with 350 μ l phenol/chloroform/isoamyl alcohol (50:50:1) plus 0.1% 8-hydroxychloroquinone. RNA samples were ethanol precipitated, dissolved in 20 μ l urea sample buffer, resolved on a 7 M urea 8% polyacrylamide gel, and visualized by neutral silver staining. Apparent electrophoretic mobility was compared with controls of known specificity.

